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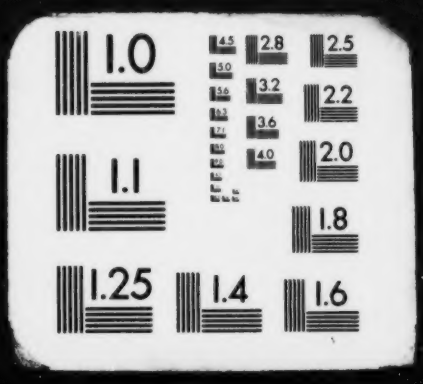

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Research and Development

Interlaboratory Evaluation of Microcosm Research

Proceedings of the Workshop

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16. ABSTRACT

This workshop, held at Athens, Georgia, on 18-19 September 1979, was to aid the Office of Environmental Processes and Effects Research (OEPER) in evaluating and integrating microcosm research in the OEPER environmental research laboratories. Participants discussed the design, advantages, and limitations of microcosms; the value and potential role of microcosms in the regulatory decision-making process; the identification of priority and long-term research needs to refine microcosm methodology as a data source for hazard and risk assessments; and the creation of a microcosm technology committee to assist OEPER in coordinating and integrating microcosm research. These proceedings consist of recommendations made by special task groups on each of the major topics discussed.

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INTERLABORATORY EVALUATION OF MICROCOSM RESEARCH: PROCEEDINGS OF THE WORKSHOP

Athens, Georgia
September 18-19, 1979

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FOREWORD

Microcosm research is one of the major components of the OEPER research program and is being carried out in six of the seven laboratories of this office. It is supported through various appropriations including TSCA and FIFRA. Microcosm research is also being actively pursued in many non-EPA laboratories. Although microcosms represent a potentially powerful tool for predicting transport, fate, and effects of toxic chemicals in the environment, this research tends to be somewhat controversial. There are disagreements among our own laboratories as to the use, limitations, and cost effectiveness of microcosms in the regulation of chemicals.

I believe that the six OEPER laboratories actively involved in microcosm research represent a unique capability for developing this tool for predicting the fate and effects of chemical substances in aquatic and terrestrial ecosystems and groundwater systems. However, in view of the complexities involved and huge resource requirements, it is desirable that the microcosm research in OEPER laboratories be efficiently coordinated and integrated.

This report will aid OEPER in evaluating and integrating microcosm research in the OEPER environmental research laboratories and stimulate communication between this office, active researchers, and the user program offices.

Allan Hirsch
Deputy Assistant Administrator
Office of Environmental Processes
and Effects Research

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ABSTRACT

This workshop, held at Athens, Georgia, September 18-19, 1979, was to aid the Office of Environmental Processes and Effects Research (OEPER) in evaluating and integrating microcosm research in the OEPER environmental research laboratories. Participants discussed the design, advantages, and limitations of microcosms; the value and potential role of microcosms in the regulatory decision-making process; the identification of priority and long-term research needs to refine microcosm methodology as a data source for hazard and risk assessments; and the creation of a microcosm technology committee to assist OEPER in coordinating and integrating microcosm research. These proceedings consist of recommendations made by special task groups on each of the major topics discussed.

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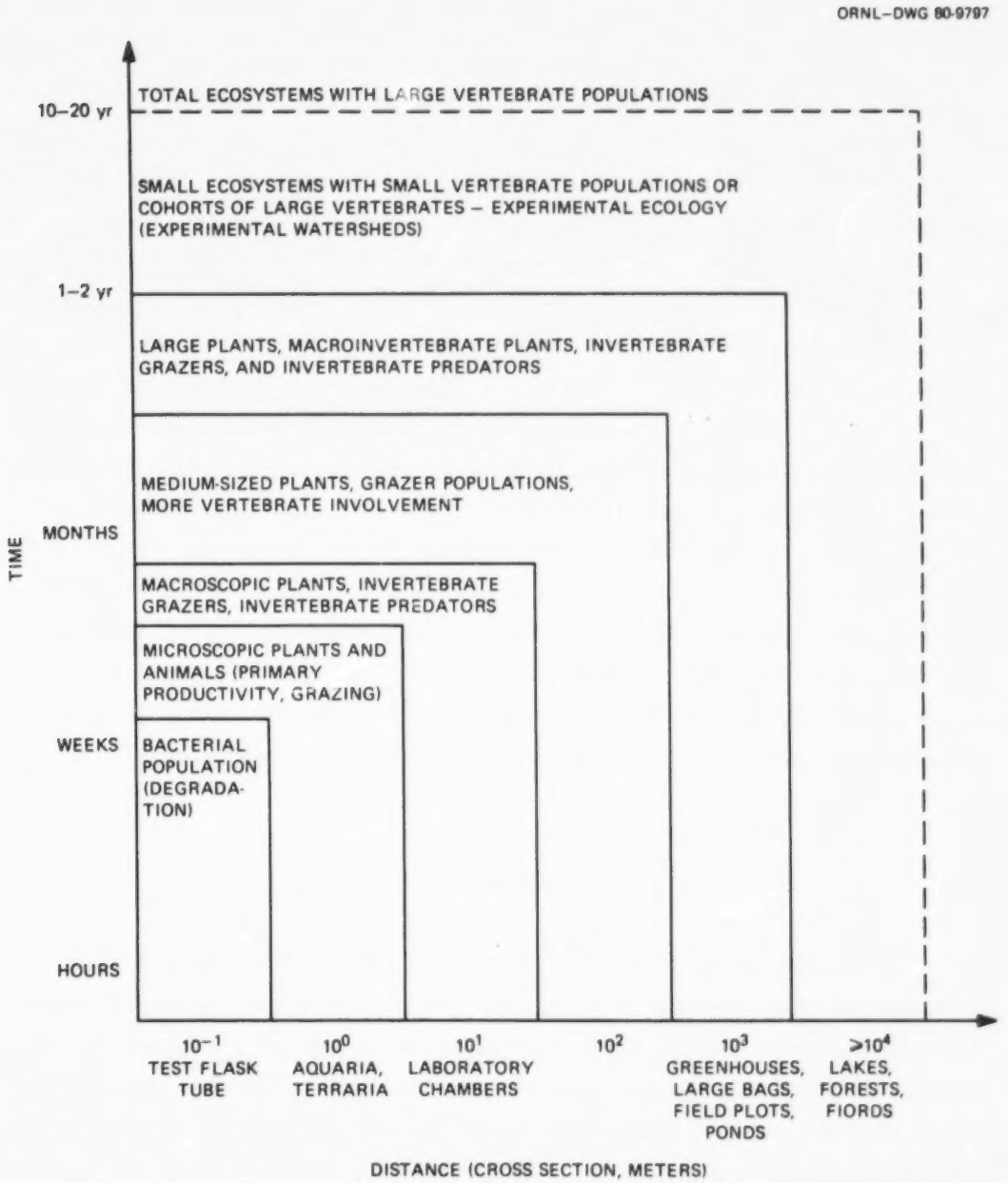
1. INTRODUCTION

On September 18-19, 1979, a workshop was held at the Sheraton Inn and Conference Center in Athens, Georgia, to discuss the current research in microcosm technology and its potential value as a tool in regulatory assessments of chemical fate and effects. The workshop was sponsored by the Office of Research and Development (ORD) and the Office of Environmental Processes and Effects Research (OEPER) of the U.S. Environmental Protection Agency (EPA). Specific topics discussed at the workshop included: the design and advantages of microcosms; the value and potential role of microcosms in the regulatory decision-making process; the identification of priority and long-term research needs to refine microcosm technology as a data source for hazard and risk assessments; and the creation of a microcosm technology committee to assist ORD and OEPER in coordinating and planning microcosm research. The workshop participants represented all major areas of microcosm research, including terrestrial, aquatic, marine, and air. Most of the U.S. research centers working with microcosms were represented. A complete list of participants and their affiliations is given in the Appendix. This report is a summary of the findings and recommendations of the workshop.

2. MICROCOSM DESIGN

The design of microcosms is dependent upon their intended uses. Interest in microcosms generally revolves around their potential to predict the behavior of selected biological and physical processes in natural ecological systems. Scalar reduction of natural ecosystems or portions of ecosystems enables investigators to include many kinds of interacting organisms and/or processes at costs consistent with the size of the microcosm. A quasi-quantitative relationship between microcosm size, complexity, and time is presented in Figure 1. In general, it is assumed that as the size of the microcosm increases, the ability to appropriately include larger fauna and flora also increases, as well as the ability to measure not only their functional but also their numerical responses. However, increases in the dimensions of experimental systems are followed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TOTAL ECOSYSTEMS WITH LARGE VERTEBRATE POPULATIONS
10–20 yr
SMALL ECOSYSTEMS WITH SMALL VERTEBRATE POPULATIONS OR COHORTS OF LARGE VERTEBRATES – EXPERIMENTAL ECOLOGY (EXPERIMENTAL WATERSHEDS)
1–2 yr
LARGE PLANTS, MACROINVERTEBRATE PLANTS, INVERTEBRATE GRAZERS, AND INVERTEBRATE PREDATORS
MEDIUM-SIZED PLANTS, GRAZER POPULATIONS, MORE VERTEBRATE INVOLVEMENT
MONTHS
TIME
MACROSCOPIC PLANTS, INVERTEBRATE GRAZERS, INVERTEBRATE PREDATORS
MICROSCOPIC PLANTS AND ANIMALS (PRIMARY PRODUCTIVITY, GRAZING)
WEEKS
BACTERIAL POPULATION (DEGRADATION)
HOURS
10^{-1} TEST FLASK TUBE
10^{0} AQUARIA, TERRARIA
10^{1} LABORATORY CHAMBERS
10^{2}
10^{3} GREENHOUSES, LARGE BAGS, FIELD PLOTS, PONDS
$>10^{4}$ LAKES, FORESTS, FIORDS
DISTANCE (CROSS SECTION, METERS)

Fig. 1. Relationships of time, scale, and biological complexity for laboratory and field microcosms and natural ecosystems.

by concomitant increases in cost. For example, a 250-ml glass container costs approximately $2.50 while a large plastic sphere capable of holding 1.9×10^9 ml costs $250,000.

Small-scale microcosms are convenient not only from the perspective of representing complex biological assemblages but also because replication is possible. Appropriate control systems can be compared using standard statistical techniques to other sets of treatment microcosms insulted by various perturbants. In addition, small-scale but complex microcosms are inexpensive to acquire and maintain. Another implicit advantage of microcosms as opposed to simulation models is the lack of any assumptions regarding a quantitative knowledge of interactions and relationships between the various physical and biological elements.

As a result of the above characteristics, microcosms can be used as a cost effective and viable tool in the management of large-scale ecological systems which are or have the potential to be perturbed by various anthropogenic substances or processes. However, there are a number of assumptions associated with extrapolating results derived from microcosms to the natural ecological system or processes being simulated. Specifically, it is assumed that the biological events observed in microcosms are either equal to or closely related to those expressed in the natural system. Rarely has this assumption been tested. As a result, development of various management strategies or regulations for natural systems based upon data derived from microcosms will always be subject to question until such assumptions are proved.

To summarize, microcosms do possess some characteristics which appear from a descriptive, qualitative point of view to be similar to those occurring in natural systems. Applications of perturbants to such systems and the ensuing results may be correlated, within the time frame of the experiments and geographic location, to those realized within natural systems which have been similarly perturbed. However, the researcher has the responsibility of demonstrating that such a correlation can, in fact, be made. Finally, it was recognized that certain fundamental characteristics of microcosms such as scale, complexity, and replicability are directly related to fabrication, operation, and maintenance costs.

The application of microcosms to the regulatory decision-making process is limited by their design and attainment of certain characteristics which allow them to produce suitable results. Some of the more important of these qualities include:

1. Microcosms used either for screening or for detailed simulation of a particular ecosystem must exhibit demonstrable and measurable ecosystem-level and community-level processes. This is necessary in order for them to be used with any assurance in predictions of ecosystem-level partitioning and biotransformation of chemicals and their bioactive residues, or in predictions of effects on other ecosystem-level attributes such as nutrient cycling, community production and respiration, predator-prey interactions, and shifts in species dominance or successional patterns.
2. The results of microcosm tests must be reproducible. Industry must be assured impartial consideration of each chemical tested. Regulatory agencies must be able to compare results of one test with another, much as the "benchmark" physical and chemical studies are used in pesticide registration.
3. Microcosms must be realistic. However, the degree of realism, or the ability of microcosm results to be extrapolated to the environment, varies depending on whether the microcosm is used for screening or for detailed simulation of a particular ecosystem. Screening tools need only display ecosystem-level interactions and need not necessarily have a physical world analog, since additional testing will follow if adverse effects are observed. Consequently, screening tools must be sensitive enough to perturbations that they will trigger additional testing, when appropriate, while still being robust enough that controls will be reproducible.

A microcosm which is designed to simulate a specific ecosystem and used to quantatively estimate the effects of potentially harmful chemicals should have real world analogs. This is required to facilitate system validation and extrapolation of results in detailed environmental assessments.

3. REGULATORY OPPORTUNITIES

3.1 REGULATORY NEEDS

There are currently immediate needs for microcosm protocols that can couple a particular ecosystem with concentration and worst-case-situation data to assist regulatory agencies in assessing the environmental fate and effects of chemicals. Environmental assessments of chemicals are presently being conducted under mandates of the Toxic Substances Control Act, the Federal Insecticide, Fungicide, and Rodenticide Act, the Federal Water Pollution Control Act, and the National Environmental Protection Act. There are currently several uses for microcosms in filling regulatory needs:

1. Microcosms are needed as screening tools to aid industry and government in the identification of those compartments of the environment with the greatest risk from the marketing and use of chemicals, thereby directing further research efforts — i.e., a "go — no go" decision in the development of a promising chemical and the selection and initiation of appropriate environmental studies.
2. Microcosms may be used to validate environmental assessment decision-making schemes that do not presently require the submission of microcosm data for every chemical. The utility of microcosm data could be measured against predictions by mathematical models of environmental fate and effects, which depend largely on physical and chemical parameters and structure-activity relationships. Thus, the flexibility of the decision maker to substitute one type of data for another, to require certain data, and to evaluate the relative degree of certainty for decisions relying on each data base could be determined. It is also probable that microcosm data could serve to fine tune mathematical models to assure better agreement among predictions from different data bases at the initial environmental assessment level.
3. Microcosms may be used to provide quality control and to supplement data submitted by industry. Again in this case screening microcosms are viewed as the most feasible technique since they can evaluate the larger number of chemicals requiring additional environmental data with lower associated manpower, time, and cost requirements.

4. Larger microcosms that simulate specific ecosystems, such as aquatic models (Perez-type at Narragansett) and terrestrial models (Corvallis-type), will be useful for later detailed environmental assessments of those chemicals identified as having potentially adverse environmental properties but which have equally great social and economic benefits. These larger microcosm systems, the results of which are more easily extrapolated to physical environmental analogs, may be used to devise mitigation procedures or to serve as the backbone either for regulatory controls on the use of the chemicals or for their withdrawal from the marketplace.

3.2 POTENTIAL REGULATORY APPLICATIONS

Most environmental regulatory actions require in some form a determination of risk or impact. Tools available for making such determinations include laboratory testing, mathematical modeling, field monitoring, and knowledge derived from previous case experience. In the case of EPA's chemical registration and premanufacture notification programs (Federal Insecticide, Fungicide, and Rodenticide Act and Toxic Substances Control Act, respectively) field data are generally unavailable, so the burden must be borne by laboratory tests (physical analog models) and predictive (mathematical) models. Most available laboratory test methods today are highly simplistic in purpose and utility. This is advantageous in that it maximizes the feasibility and unequivocal characterization of physical and biological components and attributes of ecosystems, but it is disadvantageous in that it ignores or presumes adequate understanding of the interactions between these components and attributes.

Microcosms (multicompartmental laboratory test systems) are needed in regulatory programs as part of the array of available testing protocols and as support for the development and validation of mathematical models. The range of possible uses for microcosms in a regulatory program context is extremely broad for two reasons. First, presently available technology for chemical testing and assessment demonstrates many gaps in scientific logic and empirical tools. Second, the state of microcosm technology is still embryonic and therefore holds promise of yet undetermined benefits.

The potential for such benefits arises largely from the fact that multicompartmental laboratory test systems have attributes that simply do not exist in single compartment systems. For example, the opportunity to observe chemical behavior at the water-sediment interface or in interstitial waters is provided only in a multicompartmental system. Similarly, the effects of chemicals on phenomena such as competition or other interspecific interactions, and therefore on community succession and resultant community structure, are unobservable in presently available single-species test systems.

It would be a mistake to judge the regulatory usefulness of microcosms on the basis of presently available systems, which are largely not yet ready for application and have been developed to address only some of the conceivable objectives of such systems. It would also be a mistake to set unattainable criteria for applying microcosms in a regulatory program, such as, for example, requiring quantitative proof that microcosms produce data that fully reflect the behavior of natural systems through mechanisms that are totally understood; no other test systems available today meet such criteria.

4. RESEARCH RECOMMENDATIONS

Recommended research was separated into two sections: priority research and long-term research. The priority research section was initially characterized as short-term objectives of an integrated microcosm program, but it was realized that some high priority items could not be accomplished in the short term. The distinction between short-term and long-term research is somewhat artificial since all the recommendations taken together actually form a continuum, but such a separation is necessary to identify the research most needed to assist the regulatory agencies in assessing the environmental fate and effects of chemicals. All suggested research was considered contributory to the advance of the state of the art of microcosm technology and the regulatory decision-making process.

Current priorities in regulatory program needs regarding the development of microcosms are perceived as follows:

1. Screening microcosms that, along with other tests and information (e.g., exposure and structure-activity considerations), can help determine the need and direction of further testing of the toxic hazard and/or environmental fate of a chemical.
2. Microcosms designed to simulate a specific ecosystem or subsystem that are considered as advanced predictive and confirmative tests. These systems can be undertaken when more detailed characterization of the fate and/or ecological effects of a chemical is considered to be warranted.
3. Microcosms or multicompartmental systems that can be used in a research context as tools to improve understanding of the ecological mechanisms that mediate chemical impact. These systems may or may not be used to routinely test chemicals for their environmental fate and effects.

All of these microcosms can be experimentally manipulated to aid in the development or validation of mathematical models by identifying key environmental variables and the sensitivity of model behavior to them.

4.1 PRIORITY RESEARCH

Microcosms provide unique opportunities to pursue ecological research on the environmental fate and effects of chemical substances. However, standardized uses of microcosms for routinely testing chemicals for the regulatory decision-making process are not available at this time. Therefore, current research should emphasize the refinement of microcosm methodology so that it may be used as a standard technique in the regulatory decision-making process. This refinement involves three related tasks: (1) development of streamlined testing protocols, (2) interlaboratory testing of protocols, and (3) protocol validation. Protocol development must precede interlaboratory testing, while validation could occur concurrently with protocol development and interlaboratory testing.

4.1.1 Protocol Development

Testing protocols using microcosms are needed for the implementation of the Toxic Substances Control Act. Testing protocols will be the foundation of test standards, which will become the regulations required under Section 4(a) of the Toxic Substances Control Act. They will have the force of the law and therefore will be subject to vigorous requirements for format, content, and language. Thus, testing protocols are essential to the development of consistent, defensible standards. Not only must a protocol include the conditions and procedures which need to be followed exactly for successful and replicable test performance, but a protocol must also state scientific importance, scope, and limitations of the test.

Microcosm protocols are needed for a wide spectrum of ecosystems and subsystems, especially those systems with the most social relevance. There is a particular need for screening microcosms. These microcosms may contain soil or sediment, water, microbes, invertebrates, and/or plants. They may be synthetic systems of artifically assembled components not designed to simulate a specific ecosystem, although they could consist of cores excised from a particular system. An early qualitative determination of a chemical's probable fate and effects could be made. Screening microcosms appear to be of greatest value immediately following analyses with fate models. In this position, such microcosms could confirm predictions of the fate model, and perhaps become a substitute for several single-component tests. Most important, screening microcosms could reduce both the time and cost of subsequent testing by limiting and directing the type and number of followup tests. Thus, screening microcosms could expedite the risk assessment process.

Prior to protocol documentation, microcosms need to be evaluated for their potential use in testing protocols. Hopefully, this will permit protocols to be developed more rapidly. A set of criteria to aid in this evaluation is suggested, stated in the form of a series of questions. These include, but should not be limited to, the following:

1. What specific hypothesis(es) is the microcosm being developed to test?
2. What specific attribute(s) will be used to measure the fate and environmental effects of a chemical substance?

3. How amenable is the microcosm to incorporation into a laboratory testing protocol that could be widely used by various laboratories? Has the microcosm been optimally scaled down for incorporation into a laboratory test system? Has the microcosm retained a significant amount of its ecological realism?
4. How representative are the system components compared to other possible selections of components?
5. Are the system components readily available? Can they be easily cultured?
6. How well does the test system conform to the criteria for test systems outlined by the Toxic Substances Control Act?
7. What is the basis for extrapolation of results from the system to the environment?

The order of presenting these questions is not important. The goal of this process should be to produce a definitive description of and a rationale for the test procedure.

4.1.2 Interlaboratory Testing

Interlaboratory testing, i.e., round robin testing, must follow the development of all protocols. Properly designed round robin tests permit the accuracy and precision of a testing procedure to be determined. Knowing the reproducibility of results for a promulgated test standard gives the regulatory agencies more confidence in using test results in the risk assessment and decision-making processes. Interlaboratory testing can help identify problems not observed during protocol development and allows improvements to be made in the procedures. In addition, the cost (or range of costs) of a protocol can be determined. This will allow regulatory agencies to evaluate the cost effectiveness of their testing procedures.

4.1.3 Protocol Validation

Microcosms are currently used as research models of natural systems, as aids in determining chemical fate and effects in the environment, and as methods to validate the applicability of mathematical models to natural

systems. They have been recommended for testing chemicals routinely for the regulatory decision-making process. However, if these systems are to be used to their fullest capacity, their validity as predictive tools, both separately and together, needs to be evaluated. Therefore, priority research to test the validity of these systems is recommended.

A systematic research effort should be initiated to evaluate the veracity with which microcosms exhibit those effects observed in field situations. An effort should be made to identify those perturbed ecosystems for which a sufficient amount of background information is available to permit characterization of the changes produced by specific pollutants or environmental factors. Studies should be conducted to determine the extent to which microcosms of varying levels of complexity exhibit the effects observed in the field when subjected to the same perturbing factors. Accordingly, emphasis should be placed on the following research areas:

1. Scale considerations: As research questions are formulated for which a microcosm approach is necessitated, the design of the microcosm will be greatly facilitated if background information on scale is available. The conditions should be established, including the size, that will permit the persistence of a particular trophic level, trophic interaction, or system component in the microcosm. For example, under most appropriate environmental conditions fish populations cannot be maintained in closed microcosms on natural, system-derived food unless the microcosm size approaches that available in large outdoor models. Research is needed to develop information on the effect of scale, particularly as it applies to the design of microcosm systems for both fate and toxicity studies.
2. Functional and structural parameters: Research is needed to develop and assess structural and functional parameters for comparing a microcosm to similar real world environments and to other microcosms. Functional parameters such as dissolved oxygen patterns, nutrient cycling, production-to-respiration ratios, biomass, and heterotrophic activity should be evaluated for their potential as indicators for comparing the activities of a microcosm to activities found in various natural settings. Structural parameters such as species composition,

trophic structure, and population dynamics should be investigated in a similar manner.

4.2 LONG-TERM RESEARCH

Many of the research initiatives believed to be needed are currently under way but still in early stages of development. Ongoing work in microcosm research has shown that various techniques are sensitive means of predicting the environmental fate of chemicals, but only limited progress has been made in applying existing techniques to the prediction of chemical effects. The use of relatively complex microcosms as toxicological tools is conceptually appealing because of the inherent integration of direct and indirect effects of toxic chemicals.

Preliminary efforts by various groups seem to fall into three categories or philosophies of microcosm development: (1) artificial assemblages of biota that represent functional groups; (2) natural biotic communities that are studied in situ; and (3) natural communities that are removed to the laboratory. As yet, these approaches have not been tested sufficiently to judge their respective potentials as toxicological tools.

Many kinds of information needed as a basis for further research initiatives will be obtained from this ongoing work during the next several years. However, there is a continuing need for long-term attention to the items addressed in this section.

4.2.1 Data Bases for Comparative Ecosystem Toxicology

Different kinds of ecosystems, like different kinds of organisms, will vary in their sensitivity to a particular stress. To relate the dose-response curve for one test species to a broader range of organisms requires a knowledge of the relative sensitivities of organisms. Likewise, the response of one ecosystem (natural or laboratory) will be of general predictive value only if we know the relative sensitivites of a broad range of ecosystems. There are a number of substances (e.g., certain trace elements, certain pesticides, and No. 2 fuel oil) for which

dose-response observations have been made in a variety of natural ecosystems. Such data should be compiled for several substances to provide frequency distribution curves of ecosystem sensitivity, against which the sensitivity of particular microcosm tests could be compared. The construction of such data bases represents an empirical approach to "calibrating" microcosm results for general predictions of safe exposure levels in nature.

An additional area of potential need is the identification and analysis of long-term monitoring data for ecological parameters. These data bases would aid in identifying historical trends and in determining the amount of natural variability in ecological attributes. A background would be provided against which more informed risk assessments can be made using microcosm results.

The development of these data bases will not only be useful from a comparative viewpoint, but will also aid in the validation process. These data bases can be constructed by using information from the literature and from specifically designed experiments. For example, the data bases could include information generated by the following comparisons:

1. Relate data from uncontrolled perturbations of natural ecosystems (e.g., Kepone in the James River) to microcosms of varying scale and complexity designed to simulate either a process or subsection of the perturbed natural system. The investigator would attempt to establish qualitative and quantitative relationships between the two systems for long and short time intervals.
2. Perform controlled perturbation experiments in the field where possible and in laboratory microcosms of varying scale and complexity to establish relationships of state variable dynamics between both systems.
3. Perform 1 and 2 above in different geographic locations for purposes of identifying the ecosystem independent and dependent variables and/or processes.
4. Identify the variables and/or processes of interest with respect to sensitivity and relevance to definition of system state.

4.2.2 Test Methods for Interspecific Interactions

To adequately fulfill the mandate of the Toxic Substances Control Act, laboratory test systems must be developed for predicting the effects of chemicals on those ecological phenomena that are indicative of interspecific interactions and community structure (that is, attributes which can be measured only at levels of biological organization above the population and below the ecosystem). Recent work has shown that indices of community structure and possibly of function are at least as sensitive as the "most sensitive species" approach. Attributes must be identified which represent competitive, plant-herbivore, predator-prey, host-parasite, and mutualistic relationships among species. In addition, community-level phenomena must include competition within functional groups, structural parameters, successional relationships, food chains, and food webs. There is also a need for better microcosm testing and data interpretation techniques to predict biomagnification of substances under relatively realistic steady-state conditions.

Inclusion of such laboratory test systems in the risk assessment process would impart more realism and a more integrated or holistic character to this process. The integration of multiple direct effects and indirect effects on interspecific relationships by the microcosm approach moves us many steps closer to projecting impacts on resources that have real or perceived value (e.g., fisheries). The formulation of a standard protocol for each ecological phenomenon identified is perceived as too narrow in scope. For each attribute, development of a set of protocols, each designed for a particular ecosystem type, is envisioned. For example, at the very minimum, test systems for the terrestrial, freshwater, and marine environments are needed.

4.2.3 Identification of Holistic Ecosystem Attributes

Easily measured indicators of ecosystem-level effects in microcosms must be found. Holistic or integrative attributes of ecosystems (such as energy flow, nutrient cycling, and homeostasis) are being investigated theoretically, and these studies have begun to appear in the open literature. One fact that emerges is the dependence of many of these indices

upon the same data base, namely, the flow network among the constituent compartments of the ecosystem. For example, Finn's cycling index, Mulholland's conditional entropy, May's connectivity, and Ulanowicz's ascendancy are all parameters calculated from the network of flows between system compartments. Currently, there is little agreement as to whether such attributes are clearly identifiable or useful as parameters to be evaluated in the assessment of chemical hazard. The development of practical techniques for measuring these theoretically derived attributes is recommended. In addition, ecological principles and theory need to be applied and tested in microcosms to identify the holistic attributes that are the most sensitive indicators of change in structure or function of various ecosystems. The effort to search for and validate the use of holistic attributes should receive increased attention. At the least, it will help to refine our theoretical understanding of ecology.

4.2.4 Development of Criteria for Test System Performance

Since the intended application of microcosm test results is to be an integral part of the overall risk assessment, the performance of a developed test must be quantified in terms of the goals for which it was designed, its internal variance, and its simulation of natural variance. Following such evaluation of test performance, appropriate statistical design and criteria of performance can be established prior to routine use of the test system. Statistical inferences about differences between stressed and unstressed (treated vs control) conditions are founded in probability theory, and the validity of test results, in a regulatory sense, depends on the attention paid to this aspect.

No general criteria exist for assessing what constitutes a successful microcosm. Such evaluations, of course, depend upon the purpose for which a test study is developed. In cases where microcosms are designed to closely simulate a natural system, the importance of variability in that natural system cannot be ignored. For example, if a microcosm is designed to study the fate and effects of an organic chemical, realistic results might require that the microcosm be validated by comparing the rates of major pathways, identifying major degradation products, assessing

steady-state bioaccumulation ratios, and/or examining one or more exotoxicological effects in the microcosm and the natural system.

On the other hand, microcosms designed as screening tests might in principle be less rigorously referenced to a given natural system, provided the necessary complexity of biological interaction is included. A screening microcosm could supply guidance to further testing of a chemical. For example, screening data could indicate which ecosystem components act as reservoirs and which effects require more detailed testing. In addition, predictions of media partitioning models that are based on simple physical and chemical properties can be evaluated and confirmed. Here the validation might be of a more general nature and might include the assurance that results seen are similar to what would be found in natural systems. The information base is not available to establish criteria for the degree of similarity that should be expected for either simulation microcosms or screening microcosms. Experimentation and attempts at such validation should be continued on a long-term basis so that appropriate criteria can be formulated.

4.2.5 Stochasticity and Non-Steady-State Dynamics

Data taken from laboratory microcosms usually are in the form of a time series for each compartment of interest. Often the series are compared in the original form, a procedure which on occasion could result in misleading conclusions. For example, the history of nitrate concentrations in two identical microcosms may exhibit similar form, but be shifted in phase. When compared at any one instance, the values are markedly different. Analysis of the variance applied to the primitive series would likely infer that the two systems do not replicate. Calculation of the autocorrelation function or power spectrum function, however, eliminates the effects of phasing. Appropriate statistical tests carried out upon these transformations might reveal that the time dynamics are essentially the same.

Similarly, time series analysis presents one of the few methodologies applicable to non-steady-state dynamics in microcosms. Here one is reminded of the classical analysis of electrical engineering. Single

frequency input to a system results in a spectrum of outputs which serves to characterize the response of the system. In like manner the investigator could sinusoidally vary the inputs and stresses to his system one by one. The resultant spectra of the compartments should characterize each response to the input being varied. Furthermore, cross-correlation of compartment series can be used to identify the interactions among the compartments of the system. Comparison of cross-correlation spectra between control and perturbed microcosms should allow one to focus upon those interactions most sensitive to the chemical substance under study.

Thus, while present microcosm studies are usually executed under constant laboratory environmental conditions, much more information could be obtained from model systems by temporally varying the inputs and environment in a controlled manner. Elucidation of the non-steady-state dynamics of microcosms and of their responses to chemical perturbations should, therefore, be pursued by systematically varying laboratory conditions. The judicious use of time series analysis appears to be necessary in extracting the maximum possible information from non-steady-state laboratory ecosystems.

5. INTEGRATION OF OEPER MICROCOSM PROGRAM

5.1 INTRODUCTION

It may be desirable to have some manner of formal integration of the OEPER microcosm program to carry out the following functions:

1. reduce or avoid redundance in systems development, while attempting to establish appropriate commensuration (especially between media and among media) at the same level of testing function (e.g., screening, system validation, and monitoring support);
2. establish a sense of continuity and appropriate common goals, so that necessary conjunction of research is achieved;
3. establish a mechanism for communication between researchers and program office staff (e.g., Water Quality and the Office of Pesticides and Toxic Substances), extending that communication to the scientific community through a newsletter; and

4. establish an internal peer review mechanism regarding, for example, interlaboratory testing of protocols or innovative technology.

A substantial part of these functions can be met by semiannual meetings of the involved researchers and peers in the appropriate user offices. Indeed, it would be counterproductive to add additional program reviews, extra meetings, and more paperwork. If agreement can be reached among researchers on the chemicals to be used in protocol development and round robin testing, then a single, cost effective purchase of radiolabeled material can be shared. If criteria for operations, structure, and function are agreed upon, then the products and outputs are more likely to be of utility to the program offices.

Thus, the thrust of this integrative program is to facilitate (not constrain) microcosm technology developments within limited resources (particularly of money and staff time). It is important to keep in mind that this element of the Public Health Initiative, substantially expanding ORD resources in microcosm technology, was sold to Congress as an integrated package for exposure assessment; it is not a blank check. Hence, ORD management has a responsibility to see that laboratory products fit within the mandate of the legislation. Therefore, it is proposed that a committee be established to carry out the aforementioned functions within that management framework. This group would be called the Microcosm Technology Committee (MTC).

5.2 COMMITTEE STRUCTURE

The MTC would consist of all active OEPER researchers (at least one representative from each laboratory carrying on this work) and representatives of the user program offices. This group would meet annually or, at most, semiannually to review progress, improve communication, and establish goals or criteria as desirable. The group would gather under the management and auspices of the Deputy Administrative Assistant (DAA) of OEPER.

5.3 COMMITTEE FUNCTIONS

The MTC would fulfill several functions, including providing for the following:

1. Research efficacy. An annual review, held early enough to provide ample time for input to the research committees, would provide a forum for working out the most efficient and effective means of organizing laboratory and contract research. This would provide an opportunity for workers in the same media to get together (which might otherwise be unlikely) and for those working toward the same ends (e.g., screening tests) to adjust their techniques to fill voids.
2. Continuity. There is no guarantee that a given researcher or laboratory program will necessarily continue, but the MTC would allow new researchers to develop the close contacts vital to effective research. Furthermore, it is expected that some common goals and directions will evolve from these meetings that will assist each researcher in doing the job, provide the program offices with better understanding of the limitations and potential of microcosm technology, and allow researchers to discuss the needs of each program office.
3. Communication. Among the most important functions of the committee will be the direct communication links that it establishes between the researchers, program offices, and ORD management. It is proposed that a newsletter, published by contract, be undertaken immediately.
4. Internal peer review. Since interlaboratory testing and validations of microcosm technology are likely to be expensive and more time-consuming than simple toxicity assays or chemical fate tests, it is proposed that the MTC serve as a clearinghouse or peer review group to coordinate this function according to accepted criteria. Such a function would assist management and the research committees in finding appropriate resources to bring products of microcosm technology to the stage of application by user offices.

5.4 COMMITTEE OPERATIONS

The group could elect a planning committee, or steering group, to organize the annual or semiannual meetings to assure that the program is likely to achieve its necessary functions. This steering committee would work with ORD management in initiating and supporting committee activities, such as the newsletter. Otherwise, the functions of the group will be maintained simply by the OEPER staff between meetings. It seems desirable for the MTC to meet at laboratories (preferably ORD) where microcosm work is ongoing and where in-depth discussions are more likely to arise regarding a particular technology. The meeting should be held annually (in spring or early summer) or, at most, semiannually at a time conducive to the greatest representation by researchers and users. Sponsorship of the meeting by the DAA/OEPER would facilitate travel and attendance.

APPENDIX

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